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Commentary

Multiple Factors in Carcinogenesis

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Before making some comments on multiple factors, I would like briefly to refer to the discussion at this conference. There has been no reference, strangely enough, to a law known as the Delaney Amendment. Asbestos is a carcinogen. It is apparently being deliberately added to certain drugs and foodstuffs by particular processing and manufacturing practices. Surely, when asbestos is introduced into foodstuffs by deliberate processing this is a clear cut contravention of the Delaney Amendment. The whole question of the relevance of the Delaney Amendment to asbestos in water is a complex legal matter. Suffice it to say that I think it is possible to develop a substantive legal argument that the deliberate discharge of carcinogens such as asbestos into water represents a contravention of the Delaney Amendment.

Now with relation to multiple factors — this is a fascinating area in which epidemiology has blazed the trail and toxicology has lagged sadly behind. I think that with the little we have heard here on the role of smoking and asbestos, and from the literature on the subject, it becomes clear that smoking does enhance the incidence of bronchogenic cancer following occupational exposure to asbestos. Similar considerations relate to uranium although, however, the question of the synergistic or other positive interactions between smoking and radon daughters appears somewhat compounded by the fact that silica dust may also be involved, and thus we appear to have possibly three factors: smoking, silica dust, and radon daughters. Now while it is true that epidemiology has been particularly helpful in demonstrating these relationships, there seems to have been less interest in following up the implications of such interactions. For instance, it is possible to postulate one of two entirely different concepts to account for the synergistic relationship between smoking and asbestos. One is an entirely nonspecific mechanism, based on the concept that smoking has nothing to do with asbestos and bronchogenic carcinoma except that it inhibits mucociliary defense mechanisms; other agents that might inhibit mucociliary defense mechanisms could probably also enhance the carcinogenic effects of asbestos. On the other hand, it is also possible that there is a very specific mechanism for this interaction and that the inhaled particles may interact chemically with certain carcinogens in cigarette smoke; or the particles may increase the uptake of carcinogens in cigarette smoke. It is also possible finally that carcinogens in tobacco smoke could sensitize tissues to the carcinogenic effect of the foreign particles.

It would appear difficult to differentiate epidemiologically between the so-called specific and nonspecific mechanism of smoke–particle interactions. Now, as I mentioned before, toxicology has lagged behind epidemiology in this area. As you know, forty years ago, Berenblum and others *(1)* demonstrated the two-stage mechanism of skin carcinogenesis in the mouse, showing that you have an initiating event which is a specific phenomenon; a very small dose of a carcinogen could induce sensitization of tissues to a carcinogenic effect which subsequently can be unmasked in a relatively nonspecific manner by a promoting agent. But apart from these ex-

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periments, which have historical interest and incidentally as far as I know, cannot be extrapolated to any tissues other than skin, I think toxicology has played only a small role in elucidating the whole question of multiple factors. To a certain extent, this is a reflection of the simplistic fixation which toxicology has found itself in as an off-shoot of pharmacology; it tends to study in a simplistic fashion direct causal relationships, and in many instances such studies bear little or no practical relationship to the realities of life. But that may be a little uncharitable. Perhaps what is far more to the point is that the toxicologist can very simply say, we know how to set up the tests; do we take a carcinogen which we are interested in studying, then pull 100,000 chemicals off the shelf and study them one after the other to see whether there are synergistic interactions?

Clearly this is impractical and clearly perhaps what toxicology should do is to follow well behind epidemiology, and when the possibility of synergistic interactions and multiple factors is raised, then toxicology should come in and try to get some impression as to the underlying basic mechanisms.

The toxicological literature in this area is pretty skimpy, but let me just mention one or two illustrations. You are familiar, perhaps, with the experiments of Sinnhuber, who was interested in finding apparent no-effect levels for aflatoxins and their induction of liver cancers in the rat *(2)*. Sinnhuber found that feeding aflatoxins at levels as low as 0.4 ppb was just about the lowest level which would induce cancer of the liver in trout*(3)*. Those who mistakenly believe the threshold concept for carcinogenesis were eager to seize this as an indication that levels below 0.4 ppb of aflatoxin were safe. The absurdity of this viewpoint was well exemplified by some subsequent studies, in which Sinhuber mixed the aflatoxin with a very small amount of hibiscus oil, a naturally occurring product; and instead of having an apparent no-effect level of 0.4 ppb, the apparent no-effect level reached 0.004 ppb. It is possible, therefore, to demonstrate massive shifts in apparent no-effect levels by slight modification of the experimental circumstances. Similarly there have been other experiments in which benzpyrene in trioctanoin has been tested on the skin of mice and apparent no-effect levels have been demonstrated. However when the benzpyrene has been tested in *n*-dodecane as a solvent, the apparent no-effect level shifted lower by several orders of magnitude.

Of interest in the field of particles are three sets of recent experiments. In the first of these experiments Saffiotti and his colleagues *(5)* were able to demonstrate very simply that when you put benzpyrene into the lungs of hamsters by direct endotracheal instillation, you don't get tumors; when you put iron particles into the lungs of hamsters by direct endotracheal instillation, you don't get tumors; but when you put mixtures of benzpyrene and iron oxide into the lungs of hamsters, you get a high incidence of bronchogenic squamous carcinomas. This is a clear indication of an interaction between particles and carcinogens. Again, one can say basically, it may well be a nonspecific effect, the iron merely holds the benzpyrene in place or prevents it being leached out rapidly and I think one can question exactly what the mechanisms are here. Perhaps an even more interesting experiment of Saffiotti was that which he did with Montesano *(6)* which I would strongly recommend you to consider deeply the implications if you are ever tempted to talk about apparent no-effect levels of carcinogens. Saffiotti took baby hamsters and primed them at birth with very low levels of dimethylnitrosamine or diethylnitrosamine (in the microgram range). Nothing happened to them: they grew beautifully and showed no sign of illness, no sign of sickness, no sign of tumors. Again one can say, here is a no-effect level of nitrosamines. However, if you take these hamsters which have primed at birth with very low levels of nitrosamines and when they reach adult life, you insufflate through their tracheas small levels of iron dust in the absence of a carcinogen, you will induce a high incidence of lower respiratory tract tumors. In other words, a small dose of a carcinogen at birth followed by the nonspecific presence of particles in the lungs of hamsters induces a high incidence of lower respiratory tract tumors.

Sid Laskin is here and I hesitate to talk about his work in front of him, but his work is so fascinating that let me just mention it briefly. Laskin's group *(7)* have been doing some very elegant studies in which they have been exposing rats for life to sulfur dioxide alone or to benzpyrene alone in aerosols. Animals exposed to sulfur dioxide alone or to benzpyrene alone to all intents and purposes get no bronchial carcinomas; however, if you expose rats to both benzpyrene and to sulfur dioxide, you get squamous bronchial cancers in the rats, and some squamous carcinomas of the forestomach. What does all this mean in relation to asbestos? When you load tissues and organs with asbestos what will this do to the ability of these organs to take up carcinogens from blood or from other tissues? In other words, load the tissues with asbestos and find what happens to the uptake and metabolism of other carcinogens such as N−nitroso compounds, also see what happens to the ability of alkylating agents or nitrosamines to alkylate DNA. What happens if you have asbestos present in tissues? Will this modify the alkylation of DNA by nitroso compunds or not? Similarly, can one demonstrate possible chemical interactions between other carcinogens and asbestos either *in vitro* and *in vivo.* Can the presence of asbestos in tissues increase the carcinogenic activity of other carcinogens? If, for instance, you have a carcinogen that is normally organotropic for the liver, and you load the pancreas of workers exposed occupationally to asbestos with this carcinogen, will you create a different pattern of organotropism? And again, can particles sensitize tissues to the activity of chemical carcinogens? Over and above natural organotropism, can you modify the incidence of the effect of a carcinogen by the presence of asbestos? Similarly, can chemicals sensitize tissues to the carcinogenic activity of asbestos?

These are questions to ask. I would venture to suggest that from an experimental standpoint there are no overwhelming difficulties in doing such studies and attempting to get such answers. I think that if we were able to get information of this kind we could to a certain extent begin to start complementing the massive quality and quantity of epidemiological data that has already developed on the role of multiple factors of asbestos carcinogenesis.

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